REMARKS

Claims 82 and 93 have been amended. Specifically, claim 82 has been amended to include the limitation that the coding sequence for the biologically active polypeptide is operably linked to a coding sequence for a signal peptide. Support for this amendment resides in the specification, for example, at pages 21 and 22. In view of the amendment to claim 82, claim 93 indirectly dependent therefrom has been amended to remove the duplicative phrase with respect to an operably linked coding sequence for a signal peptide. No new matter has been introduced by way of claim amendment.

Claims 82-84 and 87-94 are pending in the application. Reexamination and reconsideration of the claims is respectfully requested in view of the claim amendments above and the remarks below. The Examiner's comments in the Office Action are addressed below in the order set forth therein.

Telephonic Interview

Applicants wish to thank the Examiner for the helpful telephonic interview granted with Applicants' representative, Leslie Henry, on April 20, 2010. During this interview, the cited prior art was discussed, particularly with regard to the teachings of the Wong *et al.* reference and Applicants' position that the claimed invention is not obvious over the combined teachings of Stomp *et al.*, Wong *et al.*, Buzby *et al.*, Yu *et al.*, and Park *et al.* No agreement was reached as to allowable subject matter.

The Rejections of the Claims under 35 U.S.C. §103 Should Be Withdrawn

Claims 82-84 and 87-92 have been rejected as being obvious over International Patent Application Publication No. WO 99/07210 to Stomp *et al.* in view of Wong *et al.* (1992) *Plant Mol. Biol.* 20:81-93, Buzby *et al.*(1990) *Plant Cell* 2:805-814, and Stiekema *et al.* (1983) *Nucleic Acids Res.* 11:8051-8061. This rejection of the claims is respectfully traversed.

The claimed invention is directed toward stably transformed duckweed plant or nodule cultures that are transformed with one or more nucleotide sequences comprising a coding sequence for a biologically active polypeptide, an operably linked coding sequence for a signal peptide, and an operably linked 5' leader sequence, where the leader sequence consists of SEQ

ID NO:16, which sets forth the 5' leader of the *Lemna gibba* 5B RbcS gene. As shown throughout the application, SEQ ID NO:16 not only substantially increases production of recombinant polypeptides in duckweed, but also reduces the culture time needed for the plant or nodule cultures to produce such levels of polypeptides. Furthermore, there is no requirement that the RbcS 5' leader be operably linked to a sequence encoding a chloroplast transit peptide, or even a native transit peptide coding sequence, in order to achieve these marked improvements in recombinant protein production. Neither the enhanced polypeptide production nor the reduced culture times to achieve the enhanced production as disclosed in the present application would have predicted given the state of the art at the time of the present invention.

As the Examiner is aware, establishing a *prima facie* case of obviousness requires an assessment of the factual inquiries set forth in *Graham v. John Deere Co.*, 383 U.S. 1 (1966), which provides a framework for applying the statutory language of § 103 (*i.e.*, the "Graham Factors"). Under the Graham Factors, an examiner must:

1. Determine the scope and content of the prior art;
2. Ascertain the differences between the prior art and the claims at issue;
3. Resolve the level of ordinary skill in the pertinent art; and
4. Consider any relevant secondary considerations.

Recently, the Supreme Court identified seven (7) rationales for use in supporting obviousness determinations, which are consistent with *Graham*. MPEP § 2143. Regardless of the applied rationale, prior art "can be modified or combined to reject claims as *prima facie* obvious as long as there is a reasonable expectation of success." MPEP § 2143.02 I. The reasonable expectation of success is not required to be absolute (MPEP § 2143.02 II.), but must be determined at the time the invention was made (MPEP § 2143.02 III.). Thus, "evidence showing there was no reasonable expectation of success may support a conclusion of nonobviousness." MPEP § 2143.02.

With respect to Stomp *et al.*, Applicants again submit that this cited reference provides general teachings regarding methods of modifying nucleic acid molecules to enhance expression in duckweed of biologically active polypeptides. As the Examiner previously acknowledged, Stomp *et al.* does not contemplate or disclose using SEQ ID NO:16 alone to enhance expression of biologically active polypeptides.

Buzby *et al.* and Wong *et al.* are cited as bridging the gaps between Stomp *et al.* and the pending claims by guiding one of skill in the art to any and all RbcS 5' leader sequences, including SEQ ID NO:16. The Examiner points to Buzby *et al.* as disclosing the 5' leader sequence in SEQ ID NO:16 as a subsequence of the much larger upstream sequence of the *L. gibba* 5B RbcS gene, and relies on Wong *et al.* as providing the motivation to use this subsequence in the methods of Stomp *et al.* to arrive at Applicants' claimed invention. Stiekema *et al.* is relied on as teaching the *Lemna gibba* transit peptide. Applicants respectfully submit that the combined teachings of these four cited references fails to render Applicants' claimed invention obvious for all the reasons already of record and further in view of the following.

As the Patent Office has acknowledged on the record, the use of 5' leader sequences, including 5' leader sequences of RbcS genes **from the same plant species**, to enhance expression of heterologous proteins is an unpredictable art. Furthermore, the Patent Office relied on this fact to reject claims previously reading on the use of the genus of 5' leader sequences from *Lemna gibba* RbcS genes, of which there are approximately twelve members (Silverthorne *et al.* (1990) *Plant Molecular Biology* 15:49-58, of record). See the Office Action dated May 3, 2007, at pages 7-8, stating:

> Furthermore, even in the same plant of Lemna gibba, there are three RbcS genes disclosed by Buzby et al. with 5' leader sequences quite different in sequence (page 807, Figure 1). Only one of them was demonstrated as 5' leader sequence in the working examples. Silverthorne et al. (1990, Plant Molecular Biology 15:49-58) teach that there are at least [*sic*] genes in Lemna gibba (page 52, figure 1). Undue experimentation would also be required to determine whether other 5' leader sequences from RbcS genes of L. gibba can be used for the instant invention. . . .Given the breadth of the claims, lack of further guidance and addition [*sic*] working example [*sic*], **the unpredictability of the art**, undue experimentation would be required for a person skilled in the art to practice the invention in full scope. (*emphasis added*)

Applicants respectfully submit that the Patent Office cannot have two positions on this point, where it relies on unpredictability of the art to impose restrictions on the breadth of Applicants' claimed subject matter, yet dismisses that unpredictability in order to impose an obviousness rejection against the pending narrow claims. Either the art is predictable

and Applicants' broad claims drawn to the class of *Lemna gibba* RbcS 5' leaders were enabled, or the art is unpredictable and Applicants' claims narrowly drawn to the specific RbcS 5' leader set forth in SEQ ID NO:16 meet the requirements for nonobviousness.

As for the Patent Office's reliance on Wong *et al.* to provide the motivation to use Buzby *et al.*'s 5' leader sequence, Applicants respectfully point the Examiner to the comments of record. The results presented by Wong *et al.* teach one of skill in the art that the *Arabidopsis* RbcS 5' untranslated leader may enhance expression of a heterologous protein in **tobacco**, but such enhancement is dependent upon the coding sequence to which this leader sequence is attached, as well as being dependent upon whether or not the native transit peptide must be included in the expression construct to achieve enhanced expression. Where Wong *et al.* teach a 10-fold to 20-fold enhancement of expression, it is only achieved with a construct comprising **both** the RbcS 5' untranslated leader and its native transit peptide coding sequence. In short, Wong *et al.* teach that the effect of a RbcS 5' untranslated leader on heterologous protein expression is unpredictable. This fact has been acknowledged by the Patent Office, as discussed herein above.

Stiekema *et al.* teach the *Lemna gibba* transit peptide. Stomp *et al.* teaches that this transit peptide can be used to target expression of a heterologous polypeptide to the chloroplast. However, Applicants have discovered that substantial increases in recombinant protein expression can be achieved in duckweed by including the 5' leader sequence set forth in SEQ ID NO:16 in the transformation construct. This increase in protein expression coupled with the decrease in production time results in a duckweed expression system that provides for unexpectedly high increases in protein production that exceed the 20-fold increase taught by Wong *et al.* Furthermore, Applicants have discovered that this enhanced protein production in duckweed can be obtained with diverse heterologous polypeptides and without a requirement for the native RbcS transit peptide coding sequence in the expression construct. This discovery clearly is not taught or suggested by the combination of Stomp *et al.*, Wong *et al.*, Buzby *et al.*, and Stiekema *et al.*, and thus this obviousness rejection should be withdrawn on this basis alone.

In countering Applicants' arguments of record, the Examiner notes that the "instant claims do not contain limitations such that the heterologous polypeptide has to be secreted" and that the "invention as claimed does not exclude additional sequence to be present in addition to

SEQ ID NO:16 given the open language" (Office Action dated November 5, 2009, page 4, first full paragraph).

Applicants note that solely for the purposes of advancing prosecution, the amended claims now require that the claimed nucleotide sequence encoding the biologically active polypeptide contains an operably linked coding sequence for a signal peptide that provides for secretion of the operably linked polypeptide. Thus the claims no longer read on the presence of an operably linked RbcS transit peptide, which would direct expression of the heterologous polypeptide to the chloroplast. The combined teachings of Stomp *et al.*, Wong *et al.*, Buzby *et al.*, and Stiekema *et al.* fail to suggest Applicants' claimed invention. In fact, Wong *et al.* suggest that the *Arabidopsis* RbcS 5' untranslated leader "has evolved to be especially effective as a translational enhancer for the transit peptide coding sequence to which it is naturally attached" (see Wong *et al.*, page 91, col. 2, last sentence, continuing through line 3 of col. 1 on page 92). Thus, Wong *et al.* not only teach the necessity of using the native transit peptide coding sequence to achieve 10- to 20-fold increases in protein expression, but also point to a decided advantage of using a RbcS 5' untranslated leader with its native transit peptide coding sequence. Applicants respectfully submit that this advantage teaches away from Applicants' claimed invention.

In view of the foregoing remarks, Applicants respectfully submit that the presently claimed invention is not rendered obvious by the teachings of Stomp *et al.*, Wong *et al.*, Buzby *et al.*, and Stiekema *et al.* Reconsideration and withdrawal of this obviousness rejection is respectfully requested.

Claims 82-94 and 87-94 are rejected under 35 U.S.C. §103(a) as being obvious over Stomp *et al.* (1999, WO 99/07219) in view of Wong *et al.* (1992, *Plant Molecular Biology* 20:81-93), further in view of Buzby *et al.* (1990, *The Plant Cell* 2:805-814), further in view of Yu *et al.* (1995, U.S. Patent No. 5460952), further in view of Park *et al.* (1997, *The Journal of Biological Chemistry* 272:6876-6881), and further in view of Stiekema *et al.* (1993, *Nucleic Acid Research* 11:8051-8061). This rejection is respectfully traversed.

As noted above, the Stomp *et al.*, Wong *et al.*, Buzby *et al.*, and Stiekema references fail to demonstrate or even suggest that the 5' leader sequence set forth in SEQ ID NO:16 would be useful for expression of biologically active polypeptides in duckweed, particularly when used in absence of the native transit peptide coding sequence. The Yu *et al.* reference teaches a signal peptide for secretion of a protein into the media of the plant cell cultures. Park *et al.* teach that a signal peptide from the rice α-amylase polypeptide can be recognized and processed by various expression systems. However, neither of these additional references provide the teachings that the 5' leader of the RbcS 5B gene of *Lemna gibba* (SEQ ID NO:16) could be used in the absence of its transit peptide coding sequence to markedly enhance recombinant protein production in duckweed while decreasing culture time required to achieve these enhanced levels of expression. In view of foregoing remarks, as well as for all of the reasons of record, Applicants respectfully request reconsideration and withdrawal of this obviousness rejection.

Non-Statutory Obviousness-Type Double Patenting Rejections

Claims 82-84 and 87 stand rejected under the judicially created doctrine of obviousness-type double patenting as patentably indistinct from Claims 16-17 of U.S. Patent No. 6,815,184 to Stomp *et al.* (hereinafter Stomp II) in view of Buzby *et al.* (1990, *The Plant Cell* 2:805-814), and Wong *et al.* (1992, *Plant Molecular Biology* 20:81-93). This rejection is respectfully traversed.

The pending claims are directed toward stably transformed duckweed plant or nodule cultures that are transformed with one or more nucleotide sequences comprising a coding sequence for a biologically active polypeptide, an operably linked coding sequence for a signal peptide, and an operably linked 5' leader sequence, where the leader sequence consists of SEQ ID NO:16, which sets forth the 5' leader of the *Lemna minor* 5B RbcS gene. Stomp II, however, does not contemplate or disclose the use of this 5' leader sequence. Buzby *et al.* set forth SEQ ID NO:16 as a subsequence of a much larger upstream sequence of the *Lemna gibba* 5B RbcS gene, but provide no teaching whatsoever as to the use of this subsequence as a translational enhancer sequence. As noted above, Wong *et al.* teach a 10-fold to 20-fold enhancement of expression, but it is only achieved with a construct comprising **both** the RbcS 5' untranslated leader and its native transit peptide. Wong *et al.* also teach that the effect of a RbcS 5' untranslated leader on heterologous protein expression is unpredictable. Given the combined

teachings of these cited references, one of skill in the art had no expectation of success that SEQ ID NO:16 alone could be used in duckweed to significantly enhance heterologous protein production while decreasing culture time to achieve these increased production levels, particularly in the absence of the operably linked native transit peptide coding sequence. In view of the amendment above and foregoing remarks, as well as for all of the reasons of record, Applicants respectfully request reconsideration and withdrawal of this rejection.

Claims 82-84 and 87-94 remain rejected under the judicially created doctrine of obviousness-type double patenting as patentably indistinct from claims 3, 8-10, 23, and 26-29 of commonly owned U.S. Patent Application No. 10/794,615 by Dickey *et al.* This application has now issued as U.S. Patent No. 7,632,983. At which time allowable subject matter is agreed upon in the case of the present application, Applicants will timely file the required terminal disclaimer and appropriate fee to address this double-patenting rejection.

Claims 82-84 and 87 remain rejected under the judicially created doctrine of obviousness-type double patenting as patentably indistinct from claims 1-25 of U.S. Patent Application No. 11/778,480 by Stomp *et al.* (Stomp III) in view of Wong *et al.* (1992, *Plant Molecular Biology* 20:81-93) and Buzby *et al.* (1990, *The Plant Cell* 2:805-814). As noted in previous responses, Applicants believe this rejection should be a provisional rejection. As such, no response is required at this time. However, this application and the '480 application are commonly owned. Applicants therefore will consider the filing of a terminal disclaimer should allowable subject matter be agreed upon in either case and should the Examiner maintain the double-patenting rejection over the '480 application.

CONCLUSION

In view of the foregoing amendments and remarks, Applicants respectfully submit that the rejection of the claims under 35 U.S.C. §103 are overcome and that this application is in condition for allowance with the exception of the double-patenting objections. Early notice to this effect is solicited at which time Applicants will timely file the necessary terminal disclaimers to address the double-patenting rejections.

It is not believed that extensions of time are required, beyond those that may otherwise be provided for in documents accompanying this paper. However, in the event that additional extensions of time are necessary to allow consideration of this paper, such extensions are hereby petitioned under 37 CFR §1.136(a), and any fee required therefore is hereby authorized to be charged to Deposit Account No. 16-0605.

Respectfully submitted,

/leslie t. henry/

Leslie T. Henry
Registration No. 45,714

Customer No. 00826 **ALSTON & BIRD** LLP Bank of America Plaza 101 South Tryon Street, Suite 4000 Charlotte, NC 28280-4000 Tel Raleigh Office (919) 862-2200 Fax Raleigh Office (919) 862-2260	***ELECTRONICALLY FILED USING THE EFS-WEB ELECTRONIC FILING SYSTEM OF THE UNITED STATES PATENT & TRADEMARK OFFICE ON MAY 21, 2010.***

LEGAL02/31930343v1